ANESTHESIOLOGY

Chiral Pharmacokinetics and Metabolite Profile of Prolonged-release Ketamine Tablets in Healthy Human Subjects

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EDITOR'S PERSPECTIVE

What We Already Know about This Topic

- Ketamine produces anesthesia and analgesia with psychoactive side effects
- Norketamine, its primary metabolite, is oxidized to 2R,6R- and 2S,6S-hydroxynorketamine, which may have analgesic and antidepressant activity without adverse psychoactive effects
- Systemic exposure to the 2,6-hydroxynorketamines is three times more than to ketamine after intravenous infusion and five times more than to ketamine after drinking a ketamine solution

What This Article Tells Us That Is New

- The hypothesis that systemic exposure to the 2,6-hydroxynorketamines can be increased by administration of a prolonged-release ketamine dosage form was tested in a controlled, five-period, ascending-dose pharmacokinetic study in 15 healthy volunteers
- The (mean ± SD) oral bioavailabilities of S- and R-ketamine were 15 ± 8% and 19 ± 10%, respectively
- The systemic exposure to the hydroxynorketamine stereoisomers after oral administration of 40 mg of prolonged-release ketamine was 10 to 11 times that after administration of a comparable intravenous dose (5 mg)

ABSTRACT

Background: The anesthetic ketamine after intravenous dosing is nearly completely metabolized to R- and S-stereoisomers of the active norketamine (analgesic, psychoactive) and 2,6-hydroxynorketamine (potential analgesic, antidepressant) as well as the inactive dehydronorketamine. Oral administration favors the formation of 2,6-hydroxynorketamines *via* extensive presystemic metabolism. The authors hypothesized that plasma exposure to 2,6-hydroxynorketamines relative to the psychoactive ketamine is greater after prolonged-release ketamine tablets than it is after intravenous ketamine.

Methods: Pharmacokinetics of ketamine after intravenous infusion (5.0 mg) and single-dose administrations of 10, 20, 40, and 80 mg prolonged-released tablets were evaluated in 15 healthy white human subjects by means of a controlled, ascending-dose study. The stereoisomers of ketamine and metabolites were quantified in serum and urine by validated tandem mass-spectrometric assays and evaluated by noncompartmental pharmacokinetic analysis.

Results: After 40 mg prolonged-release tablets, the mean ± SD area under the concentrations–time curve ratios for 2,6-hydroxynorketamine/ketamine were 18 ± 11 (S-stereoisomers) and 30 ± 16 (R-stereoisomers) compared to 1.7 ± 0.8 and 3.1 ± 1.4 and after intravenous infusion (both $P < 0.001$). After 10 and 20 mg tablets, the R-ratios were even greater. The distribution volumes at steady state of S- and R-ketamine were 6.6 ± 2.2 and 5.6 ± 2.1 l/kg, terminal half-lives 5.2 ± 3.4 and 6.1 ± 3.1 h, and metabolic clearances 1,620 ± 380 and 1,530 ± 380 ml/min, respectively. Bioavailability of the 40 mg tablets was 15 ± 8 (S-isomer) and 19 ± 10% (R-isomer) and terminal half-life 11 ± 4 and 10 ± 4 h. About 7% of the dose was renally excreted as S-stereoisomers and 17% as R-stereoisomers.

Conclusions: Prolonged-release ketamine tablets generate a high systemic exposure to 2,6-hydroxynorketamines and might therefore be an efficient and safer pharmaceutical dosage form for treatment of patients with chronic neuropathic pain compared to intravenous infusion.

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The widely used ketamine is nearly completely metabolized to active primary and secondary stereoisomeric compounds by cytochrome P450-dependent enzmyes.[1] Ketamine and metabolites bind to multiple targets in the central nervous system, including the N-methyl-D-aspartate (NMDA) and α-amino-3-hydroxy-5-methyl-4-isoxazole-propionic acid glutamate receptors and the α7-acetylcholine receptor, and modulate synaptic pathways in brain tissue that are involved in initiation of narcosis or psychoactive reactions of the drug and in the pathogenesis of neuropathic pain or depressive disorders.[2,3] The parent R/S-ketamine (S > R) and, with lower efficacy, the primary metabolite R/S-norketamine produce rapid-onset and short-lived anesthesia and analgesia in patients with neuropathic pain and the undesired psychoactive adverse reactions. Norketamine is oxidized to inactive dehydronorketamine stereoisomers and

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12 isomeric forms, of hydroxynorketamine of which the 2R,6R- and 2S,6S-form are of pharmacologic interest for a potential clinical use of ketamine in patients with chronic neuropathic pain. Both stereoisomers initiate NMDA receptor–independent pharmacologic effects and lack the adverse psychoactive reactions associated with NMDA receptor antagonism. The 2,6-hydroxynorketamines exerted behavioral, electrophysiologic, and cellular antidepressant–related effects in mice (R > S),[4] which are questioned by recent experimental and clinical–pharmacokinetic data[5–7] and are the subject of a lively discussion.[8,9] Very recently, it was also shown that the 2R,6R-hydroxynorketamine exerts strong analgesic effects and relief of allodynia in three animal experimental models.[10] However, evidence from analgesic activity in clinical studies with patients is still in question and not well confirmed.[11] Nevertheless, 2R,6R-hydroxynorketamine could be a promising candidate for clinical development as a new drug for treatment of neuropathic pain.

A more realistic and efficient strategy to translate the recent experimental evidence into clinical practice would be to design an oral dosage form for ketamine, because the metabolic profile in humans is strongly dependent on the route of administration. After oral administration, the desired potential analgesic/antidepressant 2,6-hydroxynorketamines are additionally generated by presystemic, cytochrome P450-dependent "first-pass" biotransformation.[1,3] Thus, after intravenous infusion, the systemic exposure (area under the concentration–time curve from zero to infinity, $AUC_{0-\infty}$) to 2,6-hydroxynorketamines was about three times greater than to the parent ketamine. In contrast, after drinking a ketamine solution, systemic exposure to 2,6-hydroxynorketamines was five times higher than to the parent drug.[12]

We hypothesized that plasma exposure to the active 2,6-hydroxynorketamines can be additionally increased by treatment with a prolonged-release ketamine dosage form. The markedly prolonged release of small dosage portions from the multiunit tablet extends the exposure of ketamine to the metabolic enzymes along the "first-pass" route (entire small intestine, liver) and can contribute to formation of the desired 2,6-hydroxynorketamines. Therefore, we initiated a pharmacokinetic study with newly developed prolonged-release ketamine multiunit tablets to confirm higher systemic exposure to the potential analgesic/antidepressant 2.6-hydroxynorketamine relative to the analgesic/psychoactive ketamine than the intravenous administration currently practiced in the treatment of chronic neuropathic pain.

Materials and Methods

Ketamine Dosage Form

The prolonged-release ketamine tablets consist of sugar spheres coated with ketamine hydrochloride (multiunit pellets) surrounded by a sustained release membrane of water-insoluble ethyl cellulose polymer what are embedded in a hydrogel-forming polymer. Between 60 and 99% of ketamine is released within 8 h (United States Pharmacopeia basket method I; data on file of the producer, Develco Pharma Schweiz AG, Switzerland).[13]

Subjects

The nonconfirmative, exploratory study was performed in 15 healthy German white subjects (10 males, 5 females; age 20 to 35 yr; body mass index, 19.4 to 27.6 kg/m^2) genotyped for *SLC22A1* (National Center for Biotechnology Information [Bethesda, Maryland] rs12208357, rs55918055, rs72552763) according to the International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (Geneva, Switzerland) guideline for Good Clinical Practice, and to the regulations of the German Medicines Act. A statistical power calculation was not conducted. The sample size was arbitrarily set on the basis of technical considerations and our previous experience with a multiple-period design. The study was approved by the Independent Ethics Committee of the University Medicine of Greifswald (Greifswald, Germany) and by the German Federal Department of Drugs and Medicinal Products (Bonn, Germany) and was registered by eudract.emea.eu.int (identifier: EudraCT2014-000100-10) and www.ClinicalTrials.gov (identifier: NCT02494830).

The subjects were enrolled after obtaining their written informed consent and after confirmation of good health by documenting each subject´s medical history, performing a physical examination, and conducting routine clinical, chemical, and hematologic screenings. All subjects had negative results at the time of screening for drugs, human immunodeficiency virus, hepatitis B virus, and hepatitis C virus. Three subjects were smokers (<10 cigarettes/d), and 2 subjects were alcohol abstainers while 13 subjects drank alcohol occasionally. None was on a special diet (*e.g.*, vegetarian). Subjects were not taking medication with the exception of hormonal contraceptives (three female subjects). The other females used an alternative, safe method of birth control; none had a positive pregnancy test at any time during the study. Intake of grapefruit-containing food or beverages and poppy seed–containing products was not allowed from 14 days before and during the study. Subjects were requested to refrain from alcohol consumption during the study.

Study Protocol

Pharmacokinetics of ketamine were measured in a controlled, open-label, single-dose, five-period, ascending-dose study with at least 7 days washout between the study periods. The subjects were hospitalized 12 h before and up to 16 h after administration of the study medication. The study was performed under fasting conditions after oral administration of 10, 20, 40, and 80 mg racemic ketamine hydrochloride (Develco Pharma, Germany) and after

intravenous infusion of 5.0 mg racemic ketamine hydrochloride (Ratiopharm, Germany) diluted in 240 ml saline within 30 min. The overnight fasting period before medications lasted 10 h. Prolonged-release ketamine was orally administered with 240 ml tap water. Systolic and diastolic arterial blood pressure, heart rate, respiratory rate, and oxygen saturation were monitored with an IntelliVue MP2 (Philips Medicine Systems, Germany). Adverse events were detected both by a standardized questionnaire on tolerability and by querying the subjects at the scheduled times. Furthermore, the subjects were asked to report any adverse events immediately. To balance fluid intake during the absorption period of ketamine, drinking of water was standardized up to 5 h after beginning either the infusion or the oral administration. Within 24 h, drinking of water up to 2.5 l was allowed as individually desired. Standard lunch, tea time, and dinner were scheduled after 5, 8, and 11 h, respectively. The subjects had to eat the same individual amount of food in all study periods.

Venous blood was collected *via* an indwelling forearm cannula before beginning the intravenous infusion of ketamine and 10, 20, 30, 40, and 50 min and 1, 1.5, 2, 3, 4, 6, 8, 12, and 24 h thereafter. In the case of prolonged-release ketamine, blood was sampled before and after 0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4, 4.5, 5, 6, 7, 8, 12, 16, and 24 h *via* the indwelling cannula and after 36, 48, and 60 h by repeated venipuncture. Serum for quantification of ketamine and its metabolites was obtained by centrifugation at 2,000*g* at 4°C for 10 min. Urine was completely collected for 3 days and feces for 5 days. All samples were stored at –80°C until the quantitative drug assays of ketamine and its metabolites. The time span between blood sampling and freezing did not exceed 2.0 h. The clinical examinations were performed between July and October 2014 and the analytical and biometrical per-protocol evaluations between August and December 2014. After publication of NMDA-receptor independent antidepressant actions of ketamine metabolites,[4] availability of reference substances and development of tandem mass-spectrometric assays, we *post hoc* initiated the chiral quantitative and biometrical evaluations of ketamine and all metabolites in 2018 and 2019. The stereoisomers of ketamine, norketamine and dehydroketamine, were stable over the time as confirmed by comparing the stereoisomer concentrations with the initially measured racemate concentrations. For 2,6-hydroxynorketamines, long-term stability was confirmed for at least 3 months.

Analytical Methods

Chemicals. Propanol and acetonitrile (Chromasolv) were obtained in liquid chromatography–mass spectrometry quality from Carl Roth (Germany) and Sigma-Aldrich (Germany), respectively. Deionized water (conductance: less than or equal to 0.055 μS/cm, pH 5.0–6.0) was generated with an Astacus water system (membraPure, Germany).

The internal standards D4-ketamine and D4-norketamine as well as the stereoisomers of ketamine, norketamine and dehydronorketamine, and β-glucuronidase from *Helix pomatia* were from Sigma-Aldrich (Germany), and 2R,6R- and 2S,6S-hydroxynorketamine were kindly provided by the National Center for Advancing Translational Sciences (Rockville, Maryland). Methyl *tert*-butyl ether was from Merck (Germany). Stock solutions were prepared in acetonitrile and stored at −20°C. The working solutions were freshly prepared every week by using a mixture of water and acetonitrile (50:50, vol/vol) to dilute the stock solutions and stored at 4°C. All other chemicals were of analytical grade. The quantitative drug analysis was performed in our laboratory certified for Good Laboratory Practice.

Analysis of the Stereoisomers of Ketamine, Norketamine, Dehydronorketamine, and 2,6-Hydroxynorketamine. The analyses were performed as described previously.[14,15] In brief, 0.2 ml of the respective matrix were alkalized with 250 μl of sodium carbonate (1 + 1 dilution of a saturated solution) to liberate the free bases for better extraction. To quantify concentrations of total hydroxynorketamine (free metabolite plus glucuronide) in urine, the samples were treated with β-glucuronidase. Afterward, the samples were extracted by liquid–liquid extraction with methyl *tert*-butyl ether. After centrifugation, the supernatant was separated and evaporated under nitrogen at 40°C. Then, the residue was reconstituted in 100 μl of an acetonitrile/water mixture (40:60, vol/vol) and injected into the liquid chromatography–tandem mass spectrometry system. The system consisted of an Agilent 1100 series high-pressure liquid chromatography (Agilent Technologies, Germany) coupled to the triple quadrupole mass spectrometer API4000 QTRAP controlled by the validated Analyst 1.6 software (AB Sciex, Germany). The separation of the chiral compounds was performed in two steps due to the inability to separate the stereoisomers of 2,6-hydroxynorketamine. The chiral column CHIRAL-AGP (5 μm, 15 cm × 2 mm, Chiral Technologies, USA) set at 28°C was used to separate the R- and S-stereoisomers of ketamine, norketamine, and dehydronorketamine by using ammonium acetate buffer (10 mM; pH 7.5) and 2-propanol in the following gradient: 0 to 13 min (97% ammonium acetate buffer), 13.1 to 17 min (80% ammonium acetate buffer), and 17.1 to 25 min (97% ammonium acetate buffer). The chiral column Lux-Amylose-2 (5 μm, 150 × 4.6 mm [Phenomenex; Aschaffenburg, Germany] set at 40°C was used to separate the 2,6-hydroxynorketamine isomers by gradient elution with ammonium acetate buffer (5 mM; pH 9)/2-propanol:acetonitrile (4:1) as mobile phases in the following manner: 0 to 5 min (70% ammonium acetate buffer), 45 min (60% ammonium acetate buffer), 55 min (50% ammonium acetate buffer), and 55.1 to 60 min (70% ammonium acetate buffer).

Quality Assurance within Study Analyses. The linear ranges for serum measurements were 0.5 to 200 ng/ml for S- and R-ketamine, S- and R-norketamine, and S- and

R-dehydronorketamine and 1.0 to 200 ng/ml for 2S,6S- and 2R,6R-hydroxynorketamine (appendix 1). Interday accuracy values in terms of relative error and interday precision values in terms of relative SDs were within the accepted range of plus or minus 15%. For the lower limits of quantitation, the range may increase to plus or minus 20%. The ranges of quantitation and quality characteristics for all analytes are given in appendix 1.

Genotyping

Genotyping of *SLC22A1* was performed because ketamine is known to be an *in vitro* substrate for the human organic cation transporter 1 (OCT1).[16] Allelic discrimination was performed by using the commercial TaqMan SNP Genotyping Assays and TaqMan Genotyping-Master Mix on a QuantStudio 12K Flex Real-Time PCR System equipped with the Fast 96-well thermal-cycling block (Thermo Fisher Scientific, USA) under standard conditions. In detail, the following assays were used in a 10 μl polymerase chain reaction: C___30634096_10 (rs12208357), C___30634094_10 (rs55918055), and C__34211613 (rs72552763) (Thermo Fisher Scientific).

Outcome Measures

The primary outcome measure was the $AUC_{0-\infty}$. Secondary outcome measures were the maximum serum concentration and the time to maximum serum concentration, bioavailability, volume of distribution at steady state (V_{ss}), terminal half-life ($T_{1/2}$), total clearance (CL_{tot}), metabolic clearance (CL_M), and renal clearance (CL_R). Additional parameters measured were the cumulative amounts of ketamine and metabolites excreted into urine ($A_{e,\ urine}$) and into feces. Furthermore, adverse events were reported.

Pharmacokinetic Evaluation

Pharmacokinetic outcomes were evaluated by noncompartmental analysis. The maximum concentration and the time to maximum serum concentration were directly obtained from the measured concentration-time curves. The $AUC_{0-\infty}$ was calculated with the measured data points until the last quantifiable concentration (AUC_{0-t}) by the trapezoidal formula and extrapolated to infinity. Terminal half-life was calculated by $T_{1/2} = \ln 2/\lambda_z$. The terminal rate constant (λ_z) was derived from the terminal slope by log-linear regression analysis. After intravenous infusion, total clearance of ketamine was assessed by dose/$AUC_{0-\infty}$, CL_M by $CL_{tot} - CL_R$, and V_{ss} by dose × area under the moment curve/AUC^2. For both routes of administration, renal clearance was derived from the cumulative amounts ($A_{e,\ urine}$) over the $AUC_{0-\infty}$ of ketamine and the respective metabolite (A_e/AUC). Bioavailability was assessed by $AUC_{0-\infty}$ after oral administration × intravenous dose / $AUC_{0-\infty}$ after intravenous administration × oral dose. For pharmacokinetic evaluations, the IBM SPSS statistical package, version 22 (IBM, USA) was used.

Statistical Evaluation

Samples were presented as arithmetic mean ± SD. There were no missing data. All measured outcome data were included in the statistical evaluation without exclusion of any outliers. The nonparametric Mann–Whitney U test for evaluation of differences between the independent pharmacokinetic parameters of S- and R-ketamine and the Friedman test with Dunn's posttest for multiple intraindividual comparisons were selected for statistical analyses as used by convention with $P < 0.05$ as the level of statistical significance using the IBM SPSS statistical package, version 22 (IBM, USA) or GraphPad Prism 6.07 (GraphPad Software, USA).

Results

Pharmacokinetics of Intravenous Ketamine

Intravenous infusion of 5 mg ketamine hydrochloride resulted in mean ± SD maximum serum concentrations of S- and R-ketamine of 15.6 ± 4.5 and 16.5 ± 4.9 ng/ml, respectively (fig. 1, table 1). Both stereoisomers were rapidly distributed, with volumes of distribution at steady state of 6.6 ± 2.2 and 5.6 ± 2.1 l/kg, respectively. The systemic exposures ($AUC_{0-\infty}$) to 2,6-hydroxynorketamine stereoisomers were the highest, followed by exposure to the stereoisomers of norketamine, ketamine, and dehydronorketamine, with significant differences in the exposure to the stereoisomers of 2,6-hydroxynorketamines (R > S; $P < 0.001$) and dehydronorketamine (R > S, $P = 0.006$). The $AUC_{0-\infty}$ values of S- and R-norketamine were 1.7 ± 0.6 and 1.5 ± 0.6 times the $AUC_{0-\infty}$ of R- and S-ketamine (both $P < 0.001$), and the exposures to 2S,6S- and 2R,6R-hydroxynorketamine were 1.7 ± 0.8 and 3.1 ± 1.4 times the exposure to S- and R-ketamine, respectively (both $P < 0.001$). The $AUC_{0-\infty}$ ratio between 2R,6R-hydroxnorketamine and S-ketamine was 3.4 ± 1.0 ($P < 0.001$).

Ketamine was nearly completely eliminated by metabolic disposition. The metabolic clearances of R-ketamine and S-ketamine of 1,530 ± 380 ml/min and 1,620 ± 380 ml/min, respectively, characterized ketamine as a high-clearance drug. Terminal half-lives of S- and R-ketamine were 6.1 ± 3.1 and 5.2 ± 3.4 h. Only negligible amounts of both ketamine stereoisomers were excreted unchanged by the kidneys (both 50 ± 40 μg). On the other hand, approximately 8.0% of the dose was recovered in the urine in the form of S-metabolites (norketamine, 60 ± 20 μg; dehydronorketamine, 230 ± 50 μg; 2,6-hydroxynorketamine, 60 ± 30 μg) and approximately 20% as R-metabolites (norketamine, 50 ± 20 μg; dehydronorketamine, 280 ± 60 μg; 2,6-hydroxynorketamine, 120 ± 50 μg) with significant differences for dehydronorketamine (R > S; $P = 0.022$)

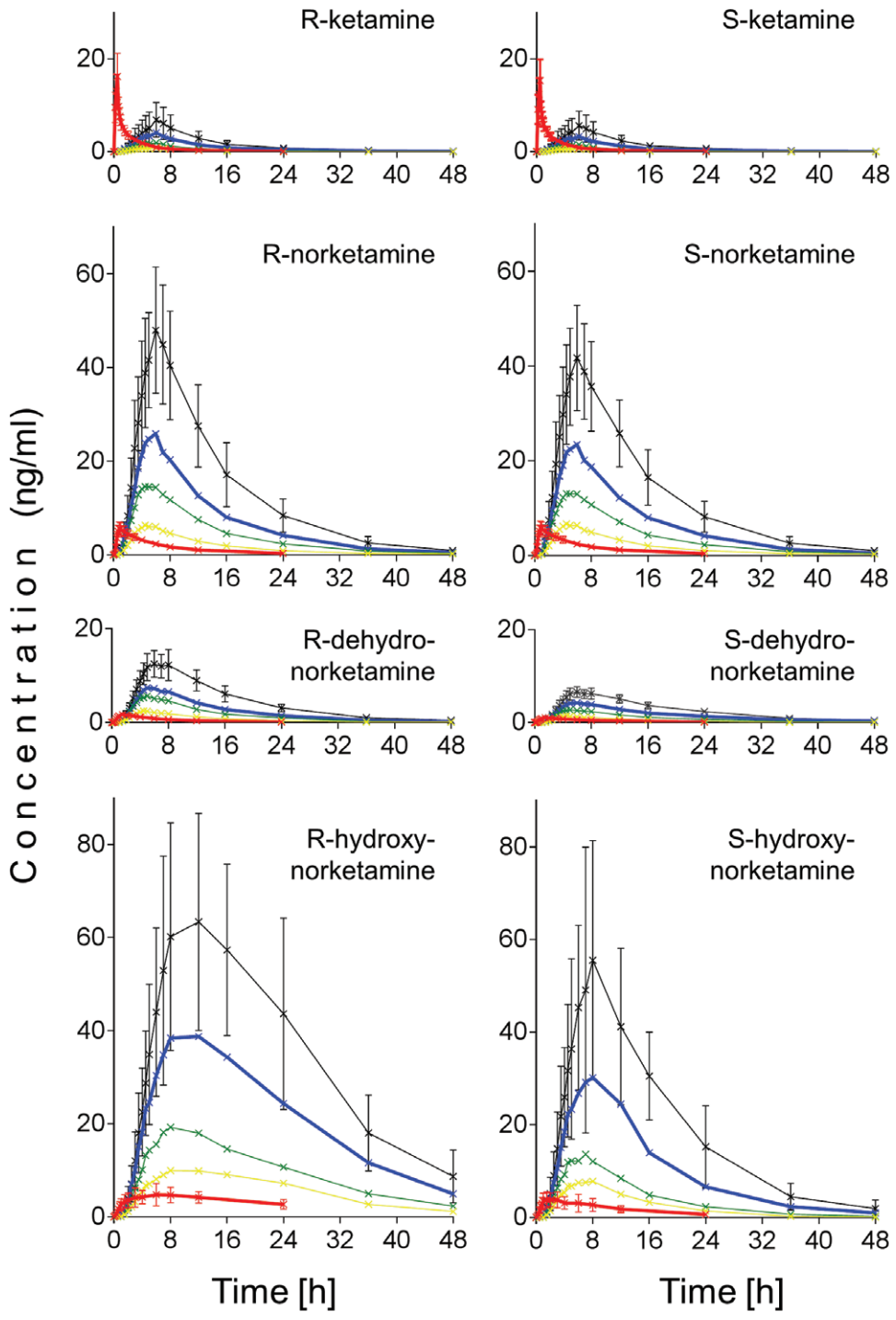

Fig. 1. Mean serum concentration–time curves of the R- and S-stereoisomers of ketamine and norketamine, dehydronorketamine, and 2,6-hydroxynorketamine after intravenous infusion of 5 mg ketamine hydrochloride solution within 30 min (*bold red lines*) and oral administration of 10 mg (*yellow lines*), 20 mg (*green lines*), 40 mg (*bold blue lines*), and 80 mg (*black lines*) prolonged-release ketamine tablets in 15 healthy subjects. SDs are given for the intravenous infusion and 80 mg tablets only.

and 2,6-hydroxynorketamine (R > S; $P < 0.001$). The 2,6-hydroxynorketamines were excreted also as glucuronides (S-form, 80 ± 40 μg; R-form, 620 ± 180 μg; $P < 0.001$). The renal clearances of S- and R-ketamine (35 ± 37 and 33 ± 35 ml/min), S- and R-norketamine (24 ± 6 and 23 ± 6 ml/min), and 2S,6S- and 2R,6R-hydroxynorketamine

Table 1. Pharmacokinetics of R- and S-ketamine after Intravenous Infusion of 5 mg Ketamine and 10, 20, 40, and 80 mg Prolonged-release Tablets

		5 mg Intravenous Infusion		10 mg Tablets		20 mg Tablets		40 mg Tablets		80 mg Tablets	
Pharmacokinetic Outcome Measures	**Unit**	**S-isomer**	**R-isomer**	**S-isomer**	**R-isomer**	**S-isomer**	**R-isomer**	**S-isomer**	**R-isomer**	**S-isomer**	**R-isomer**
Ketamine											
$AUC_{0-\infty}$	ng × h/ml	27 ± 6	29 ± 6	6 ± 47	8 ± 5	13 ± 10	18 ± 13	33 ± 19	42 ± 25	58 ± 32	72 ± 39
Maximum serum concentration	ng/ml	15.6 ± 4.5	16.5 ± 4.9	0.8 ± 0.4	1.0 ± 0.6	1.6 ± 1.1	2.0 ± 1.3	3.4 ± 2.0	4.2 ± 2.5	5.7 ± 3.1	7.0 ± 3.7
Time to maximum serum concentration	h	—	—	4.9 ± 1.3	5.1 ± 1.1	5.6 ± 0.8	5.6 ± 0.8	5.8 ± 0.9	5.9 ± 0.9	6.1 ± 0.7	6.1 ± 0.4
Bioavailability	%	—	—	12 ± 7	14 ± 9	12 ± 9	16 ± 11	15 ± 8	19 ± 10	14 ± 7	16 ± 8
Volume of distribution at steady state	l/kg	6.6 ± 2.2	5.6 ± 2.1	—	—	—	—	—	—	—	—
Terminal half-life	h	6.1 ± 3.1	5.2 ± 3.4	5.8 ± 3.2	6.0 ± 2.9	5.7 ± 2.2	7.6 ± 2.2	10.0 ± 3.9‖	11.1 ± 4.3#	8.7 ± 2.1	10.3 ± 2.9#
Cumulative amount excreted into urine	µg	50 ± 40	50 ± 40	20 ± 10	30 ± 10	50 ± 20	60 ± 20	100 ± 50	120 ± 60	180 ± 70	220 ± 80
Metabolic clearance	ml/min	1,620 ± 380	1,530 ± 380	—	—	—	—	—	—	—	—
Renal clearance	ml/min	35 ± 37	33 ± 35	71 ± 24#	71 ± 24#	75 ± 34‖	62 ± 33	57 ± 19	50 ± 19	60 ± 17§	58 ± 21§
Norketamine											
$AUC_{0-\infty}$	ng × h/ml	44 ± 12	41 ± 12	83 ± 22	80 ± 24	176 ± 83	192 ± 89	303 ± 80	322 ± 834	581 ± 187	626 ± 216
Maximum serum concentration	ng/ml	6.2 ± 1.3	6.0 ± 1.3	7.2 ± 1.6	6.7 ± 1.2	13.9 ± 3.8	15.9 ± 4.4	23.9 ± 5.3	26.3 ± 5.7	42.8 ± 10.5	49.1 ± 13.1
Time to maximum serum concentration	h	0.9 ± 0.3	0.9 ± 0.3	4.8 ± 0.9	4.7 ± 0.9	4.8 ± 0.9	4.8 ± 0.8	5.6 ± 0.6	5.7 ± 0.7	6.3 ± 0.6	6.3 ± 0.5
Terminal half-life	h	6.9 ± 2.2	6.0 ± 2.1	7.3 ± 1.2	8.4 ± 3.5	8.0 ± 2.1	7.4 ± 1.7	7.5 ± 1.4	7.9 ± 1.1	8.6 ± 2.7	7.4 ± 1.0
Cumulative amount excreted into urine	µg	60 ± 20	50 ± 20	110 ± 20	90 ± 20	230 ± 80	190 ± 70	410 ± 110	350 ± 110	800 ± 240	710 ± 240
Renal clearance	ml/min	24 ± 6	23 ± 6	23 ± 5	21 ± 8	23 ± 8	18 ± 6	23 ± 6	18 ± 4.4*	24 ± 8	20 ± 6
Dehydronorketamine											
$AUC_{0-\infty}$	ng × h/ml	8 ± 3	11 ± 4†	16 ± 7	29 ± 10‡	40 ± 14	73 ± 33‡	71 ± 10	100 ± 13‡	122 ± 23	199 ± 42‡
Maximum serum concentration	ng/ml	1.0 ± 0.4	1.8 ± 0.6‡	1.5 ± 0.5	2.6 ± 0.7‡	3.0 ± 1.1	6.1 ± 2.3‡	4.7 ± 1.3	7.9 ± 1.6‡	6.8 ± 1.2	13.4 ± 3.1‡
Time to maximum serum concentration	h	1.7 ± 0.6	1.8 ± 0.5	5.1 ± 0.7	4.8 ± 0.3	5.8 ± 1.1	5.3 ± 1.1	6.3 ± 1.3	5.8 ± 1.1	6.5 ± 1.0	6.6 ± 1.2
Terminal half-life	h	7.9 ± 4.4	4.7 ± 1.2*	11.4 ± 6.0	7.6 ± 1.7†‖	10.5 ± 2.9	8.8 ± 3.8*#	11.3 ± 2.7§	8.2 ± 1.4†#	10.3 ± 1.3	8.4 ± 2.1‡#
Cumulative amount excreted into urine	µg	230 ± 50	280 ± 60*	480 ± 140	560 ± 130	1,060 ± 290	1,150 ± 290	1,560 ± 360	2,070 ± 540*	3,250 ± 840	4,030 ± 780*
Renal clearance	ml/min	620 ± 340	440 ± 120	590 ± 240	350 ± 190†	490 ± 190	300 ± 110†‖	370 ± 82	350 ± 110	450 ± 110	350 ± 80†
2,6-Hydroxynorketamine											
$AUC_{0-\infty}$	ng ×h/ml	46 ± 28	92 ± 30‡	101 ± 33	263 ± 92‡	178 ± 87	475 ± 180‡	462 ± 194	1,050 ± 411‡	861 ± 372	1,720 ± 650‡
Maximum serum concentration	ng/ml	4.5 ± 1.7	5.2 ± 2.0	9.3 ± 4.1	11.0 ± 4.1	16.0 ± 6.5	20.3 ± 7.9	36.4 ± 15.7	43.6 ± 17.5	58.9 ± 26.7	66.1 ± 23.3*
Time to maximum serum concentration	h	2.5 ± 1.6	6.7 ± 2.3‡	6.6 ± 1.1	9.6 ± 3.5†	6.3 ± 1.3	8.4 ± 2.8†	7.2 ± 1.9	9.7 ± 2.8	7.4 ± 1.0	12.0 ± 2.9‡
Terminal half-life	h	9.9 ± 4.6	18.1 ± 6.2†	6.8 ± 1.9	10.1 ± 1.6‡§	7.4 ± 3.4	11.1 ± 3.43‡§	9.0 ± 4.6	10.3 ± 2.8*‖	8.3 ± 2.0	9.7 ± 2.1*‖
Cumulative amount excreted into urine with glucuronide	µg	80 ± 40	620 ± 180‡	150 ± 40	800 ± 290‡	500 ± 180	1,990 ± 570†	970 ± 370	4,440 ± 1,200	2,090 ± 700	9,780 ± 2,000*
Cumulative amount excreted unchanged into urine	µg	60 ± 30	120 ± 50‡	120 ± 50	230 ± 90‡	250 ± 100	420 ± 130†	710 ± 310	1,010 ± 400	1,680 ± 940	2,180 ± 900*
Renal clearance	ml/min	24 ± 10	25 ± 11	20 ± 8	15 ± 5*‖	30 ± 20	16 ± 5†‖	29 ± 14	17 ± 6	34 ± 12§	25 ± 15†

*$P < 0.05$, †$P < 0.01$, ‡$P < 0.001$, Mann–Whitney U test for comparisons between R- and S-stereoisomers; §$P < 0.05$, ‖$P < 0.01$, #$P < 0.001$, Friedmann test with Dunn's posttest for comparisons with 5 mg infusion. Exact *P* values for all Mann-Whitney U tests and Friedman and Dunn's multiple comparisons tests are provided in appendix 3.

$AUC_{0-\infty}$, area under the concentration-time curve from zero to infinity.

(24 ± 10 and 25 ± 11 ml/min) were much lower than the physiologic glomerular filtration rate in healthy subjects. In contrast, renal clearances of S- and R-dehydronorketamine (620 ± 340 and 440 ± 120 ml/min) indicated substantial tubular secretion. Renal clearances were not significantly influenced by chirality.

Pharmacokinetics of Prolonged-release Ketamine

Due to low oral bioavailability of S- and R-ketamine (15 ± 8 and 19 ± 10%), the 40 mg prolonged-release ketamine tablets showed $AUC_{0-\infty}$ values of S- and R-ketamine of 33 ± 19 and 42 ± 25 ng × h/ml, respectively, which were similar to the values after intravenous infusion of 5 mg ketamine. Therefore, we report here the pharmacokinetic measures of our 40 mg prolonged-release tablets. The differences in comparison to the other dosage forms are reported below.

Prolonged-release ketamine was very slowly released and reached maximum serum concentrations after 5.8 ± 0.9 h (S-isomer) and 5.9 ± 0.9 h (R-isomer). Due to the prolonged-release design of the dosage form, the terminal half-lives of S- and R-ketamine were 10.0 ± 3.9 h and 11.1 ± 4.3 h, respectively, nearly twice as long as after intravenous infusion (S-isomer, $P = 0.005$; R-isomer, $P < 0.001$). Only traces of the oral dose were excreted unchanged into the urine (S-isomer, 100 ± 50 μg; R-isomer, 120 ± 60 μg).

After 40 mg prolonged-release ketamine, the $AUC_{0-\infty}$ of S- and R-norketamine (303 ± 80 and 322 ± 83 ng × h/ml) and of S- and R-dehydronorketamine (71 ± 10 and 100 ± 13 ng × h/ml, $P < 0.001$) were approximately 7.0- to 9.0-fold higher compared to intravenous infusion of 5 mg ketamine. The exposure to 2R,6R-hydroxynorketamine was approximately 2.3-fold higher than to 2S,6S-hydroxynorketamine (1,050 ± 411 *vs.* 462 ± 194 ng × h/ml, $P < 0.001$). The systemic exposure to both stereoisomers was 10- to 11-fold more than after administration of a comparable intravenous dose (2R,6R-hydroxynorketamine: 1,050 ± 411 *vs.* 92 ± 30 ng × h/ml; 2S,6S-hydroxynorketamine: 462 ± 194 *vs.* 46 ± 28 ng × h/ml). The $AUC_{0-\infty}$ of S- and R-norketamine exceeded the $AUC_{0-\infty}$ of S- and R-ketamine by the factors 12 ± 5 and 10 ± 6, whereas the $AUC_{0-\infty}$-values of 2S,6S- and 2R,6R-hydroxynorketamine exceeded the values of S- and R-ketamine by the factors 18 ± 11 and 30 ± 16. The $AUC_{0-\infty}$-ratio 2R,6R-hydroxnorketamine/S-ketamine (potential analgesic/antidepressant over psychoactive exposure) was 39 ± 25 compared to 3 ± 1 after intravenous dosing (all comparisons with intravenous ketamine, $P < 0.001$).

Approximately 7% and 17% of the oral dose was excreted in the urine as S- and R-metabolites (norketamine, 410 ± 110 and 350 ± 110 μg; 2,6-hydroxynorketamine, 710 ± 310 and 1,010 ± 400 μg), with significant differences for dehydronorketamine (1,560 ± 360 and 2,070 ± 540 μg, $P = 0.011$). Only traces of ketamine and metabolites were recovered in the feces.

The pharmacokinetic results for the 10, 20, and 80 mg prolonged-release ketamine tablets were similar to the data obtained for the 40 mg dosage form. However, there was some evidence for dose-dependency in metabolic disposition even though the results were not fully consistent at all points. Significant differences were obtained in the systemic exposure to norketamine relative to ketamine and 2,6-hydroxynorketamine relative to ketamine after the higher doses (40 mg and 80 mg) compared to the low doses of 10 and 20 mg (fig. 2). At higher doses, the exposures to R- and S-norketamine relative to S- and R-ketamine and 2R,6R-hydroxynorketamine relative to R-ketamine, respectively, were significantly smaller compared to the lower doses. The exposure to 2,6-hydroxynorketamine over norketamine increased with dose in case of the S-stereoisomer but not of the R-form.

Influence of SLC22A1 Polymorphisms

The *SLC22A1* transporter polymorphisms did not influence any pharmacokinetic characteristics of the ketamine disposition.

Adverse Events of Prolonged-release Ketamine

Prolonged-release ketamine was safe and well-tolerated. Twenty-six adverse events occurred during the entire study; seven adverse events were considered by the clinical investigators not to be or unlikely to be related to administration of prolonged-release ketamine. Four events were considered possibly, 11 probably related to the study medication, and 4 adverse events were not assessable. Dizziness (8 events), headache (4 events), and palpitations (3 events) were the most frequent adverse events. The study medication did not change systolic and diastolic blood pressure, heart rate, breathing rate, and oxygen saturation in a clinically relevant manner from baseline. Serious adverse events and suspected unexpected serious adverse drug reactions did not occur.

Discussion

We gathered comprehensive data on chiral pharmacokinetics and metabolic disposition of ketamine after single oral administration of ascending doses of prolonged-release multiunit tablets in healthy human subjects. All previous attempts to quantify ketamine pharmacokinetics after oral administration were limited by inclusion of only the S-enantiomer in the data, a lack of an intravenous control study arm, the absence of chiral quantitative concentration–time data for ketamine and the active metabolites in plasma and urine, or the inaccuracy of pharmacokinetic outcome measures, *e.g.*, of the study-derived AUC_{0-t}, which must be greater than 80% of the $AUC_{0-\infty}$.[12,17–23]

Concerning the primary pharmacokinetic outcome measure of our study, we confirmed that the plasma exposure ($AUC_{0-\infty}$) to 2,6-hydroxynorketamines was up to 30 to 40 times greater (R > S) than to ketamine. These results clearly confirmed our hypothesis, that the chiral metabolic profile of ketamine after oral administration of a prolonged-release

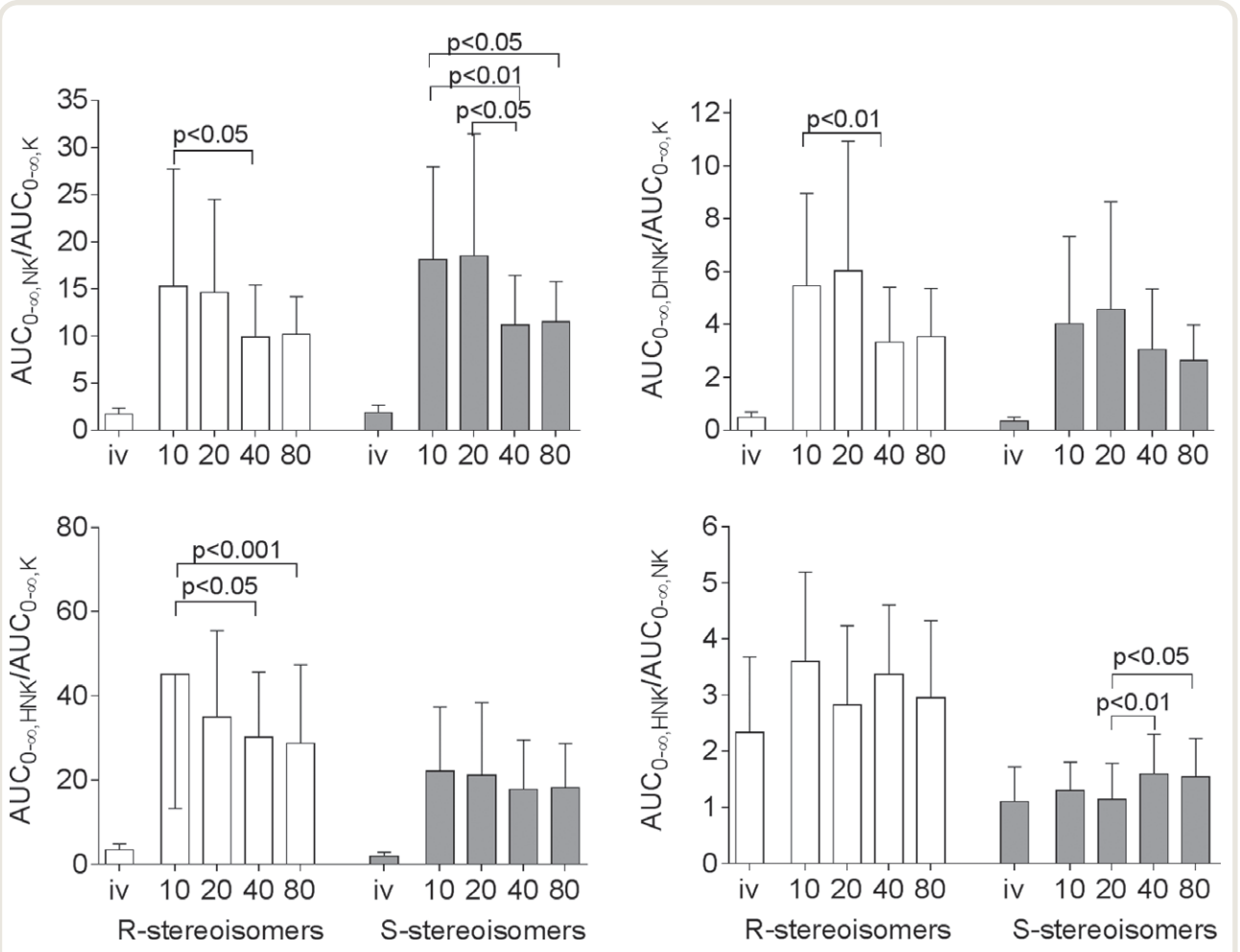

Fig. 2. Mean ± SD area under the concentration-time curve from zero to infinity ($AUC_{0-\infty}$) ratios between the R- and S-stereoisomers of ketamine ($AUC_{0-\infty,K}$), norketamine ($AUC_{0-\infty,NK}$), dehydronorketamine ($AUC_{0-\infty,DHNK}$), and 2,6-hydroxynorketamine ($AUC_{0-\infty,HNK}$) after intravenous (iv) infusion (5 mg, iv) and oral administration of 10, 20, 40, and 80 mg prolonged-release ketamine tablets in 15 healthy subjects. The *brackets* indicate significance levels for differences between the respective ratios after oral dosing to confirm potential pharmacokinetic dose dependency. After iv infusion, all $AUC_{0-\infty}$ ratios were significantly different from the ratios of all oral doses (at least $P < 0.05$) except the R- and S-hydroxynorketamine/norketamine ratios, which were only significantly different from the 40 mg tablets ($P = 0.016$ and $P = 0.022$). The exact *P* values for all Friedman and Dunn's multiple comparisons tests are provided in appendix 2.

ketamine dosage form is markedly different than after intravenous infusion, and both are different than the profile of immediate-release ketamine. Thus, Rao *et al.* found an $AUC_{0-\infty}$ ratio for racemic 2,6-hydroxynorketamine/ketamine of 5.0 ± 2.0 after drinking a 30 mg solution.[12] With regard to formation of the primary metabolite norketamine, there appears to be no marked differences between 30 mg drinking solution and 40 mg prolonged-release tablets. In contrast, hydroxylation of norketamine to the 2,6-hydroxynorketamines was more extensive after prolonged-release tablets compared to ketamine solution.[12]

Based on the $AUC_{0-\infty}$ ratio for racemic 2,6-hydroxynorketamine/norketamine (approximately 2.25), we can (at least roughly) assess a hypothetical daily dose for our prolonged-release ketamine tablets to achieve plasma concentrations of the active 2,6-hydroxynorketamine between 30 and 40 ng/ml, which were achieved in patients with drug-resistant major depression who responded to an intravenous infusion of 0.5 mg/kg ketamine.[24] Considering that the steady-state plasma concentration of norketamine after twice daily dosing of 240 mg prolonged-release ketamine ranges between approximately 200 and 300 ng/ml (fig. 2 in Glue *et al.*)[25] and the 2,6-hydroxynorketamine/norketamine ratio is about 2.25, the 2,6-hydroxynorketamine concentrations should range between 450 and 680 ng/ml.[25] The daily oral dose to achieve plasma concentrations similar to those reported in responders to intravenous therapy (30 to 40 ng/ml) should therefore be about 40 mg daily (20 mg

prolonged-release tablets twice daily) assuming dose-independency of ketamine disposition.

Ketamine is a high-clearance drug with a metabolic clearance of about 1,500 ml/min in our pharmacokinetic study, which resembles the physiologic liver blood flow in healthy subjects. Considering a plasma protein binding of 10 to 50%[3] and the low bioavailability of about 12 to 19%, pharmacokinetics and metabolic disposition of our prolonged-release tablets should be highly susceptible to changes in liver blood flow and metabolic capacity of cytochrome P-450–dependent enzymes. Therefore, clinically relevant particularities in metabolic profile and systemic exposure to ketamine and active metabolites are most likely to be expected in patients with chronic liver diseases. On the other hand, ketamine, norketamine, and 2,6-hydroxynorketamine are most likely subjected to glomerular filtration followed by tubular reabsorption (renal clearance approximately 23 to 35 ml/min) and dehydronorketamine to filtration and tubular secretion (renal clearance approximately 350 to 620 ml/min). Therefore, changes in renal function are not expected to be clinically relevant due to the small dose fractions of the pharmacologic active compounds recovered in urine and due to the low renal clearances relative to the major metabolic clearance of ketamine.

An explanation for the favored formation of 2R,6R-hydroxynorketamine after oral administration of prolonged-release ketamine compared to oral drinking solution and relative to intravenous infusion is complex and could be explained by a number of mechanisms. First, R-ketamine might be better absorbed from the gut and more extracted by the liver to be fully converted *via* R-norketamine to additional R-hydroxynorketamine. The bioavailability of R-ketamine or the R-norketamine/R-ketamine $AUC_{0-\infty}$ ratio must not necessarily be changed relative to S-ketamine. The common mechanism for intestinal absorption and hepatic uptake of ketamine could be transport *via* OCT1, which serves as an uptake carrier for organic cations in both the intestine and the liver. Ketamine is a substrate for organic cation transporters *in vitro*,[16] and it has already been shown in experiments with transfected cells that the uptake by OCT1 can be stereoselective for a chiral drug.[26] However, we could not support this transporter rationale with outcomes measured in our study; *i.e.*, there were no marked differences in the four carriers of frequent OCT1 gene polymorphisms among our 15 healthy subjects. Second, either the formation clearances of R-norketamine and/or 2R,6R-hydroxynorketamine are higher than those of the S-stereoisomers, or the elimination clearance of S-hydroxynorketamine markedly exceeds the clearance of R-hydroxynorketamine. There is some evidence that both formation and elimination clearances are different. Desta *et al.*[1] confirmed with enzyme-kinetic studies with human liver microsomes that the intrinsic formation clearance for R-hydroxynorketamine is markedly higher than for the S-stereoisomer. The conversion of norketamine to hydroxynorketamine is primarily mediated by cytochrome P4502A6, and this process is highly enantioselective and favors R-norketamine. From figure 1, it can be observed that 2R,6R-hydroxynorketamine reaches a markedly higher serum concentration, forming a short plateau, and is available longer than the S-stereoisomer. The terminal half-lives of 2R,6R-hydroxynorketamine were generally significantly longer than of the S-stereoisomer. Finally, R-ketamine may inhibit the elimination of S-ketamine.[27]

As a secondary outcome measure in our study, we demonstrated that our prolonged-release ketamine tablets are safe and well-tolerated up to doses of 80 mg ketamine hydrochloride. Dissociative/hallucinogenic disturbances (*e.g.*, lucid dreaming, nightmare, agitation, motor restlessness, confusion) or other frequent adverse reactions (e.g., tachycardia, blood pressure increase, sight disorders, hypersalivation), which are expected after intravenous administration of anesthetic (1.0 to 2.0 mg/kg, approximately 75 to 150 mg) or analgesic/antidepressant doses (0.5 mg/kg, approximately 40 mg),[28] did not occur up to the highest dose of our prolonged-release ketamine tablets. Other authors reported that ascending repeated oral dosing of prolonged-release ketamine up to 240 mg daily almost completely lacked dissociative/hallucinogenic disturbances; of 12 patients with treatment-resistant depression and anxiety, 1 subject complained of dizziness, 1 of dissociation, and 3 of headaches.[25] In healthy subjects, repeated dosing of 60 and 120 mg caused no dissociation, but headaches (three reports) and dizziness (two reports). In the 240 mg oral dose group, however, dissociation was reported by 11 subjects.[20] In summary, the risk of undesired psychoactive effects seems to be markedly lower after treatment with our prolonged-release ketamine tablets than after intravenous infusion of comparable doses commonly used in therapy of neuropathic pain and treatment-resistant depression.

Limitations of the Study

We did not include an additional study period with immediate-release ketamine because of regulatory requirements. Therefore, we cannot provide own controlled data on the meaning of ketamine absorption rate for presystemic formation of the desired 2,6-hydroxynorketamines. A second, theoretical bias might be long-term stability of the 2,6-hydroxynorketamines at –20°C, which we could confirm for 3 months only. However, instability would result in underestimation of the stereoisomer concentrations instead to the manyfold higher exposure.

Conclusions

Prolonged-release ketamine tablets generate a high systemic exposure to the potential analgesic/antidepressant 2,6-hydroxynorketamines and therefore might be an efficient and safer pharmaceutical dosage form for long-term

treatment of patients with chronic neuropathic pain compared to intravenous infusion.

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Competing Interests

Dr. Rey is an employee of Develco Pharma Schweiz AG (Pratteln, Switzerland). The other authors declare no competing interests.

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Appendix 1. Quality Control Results of the Quantitative Analysis of R- and S-ketamine, R- and S-norketamine, R- and S-dehydronorketamine, and 2R,6R- and 2S,6S-hydroxynorketamine in Serum and Urine and of R/S-ketamine and R/S-norketamine in Feces

Analyte	Matrix	Range (ng/ml)	Quality controls (ng/ml)	Precision (%)	Accuracy (%)
S-ketamine	Serum		1.5	12.7	14.2
		0.5–200	100	7.6	–1.1
			200	4.2	3.3
	Urine		2.5	7.0	1.9
		1.5–500	250	3.7	–2.9
			500	4.3	0.3
R-ketamine	Serum		1.5	6.4	6.8
		0.5–200	100	7.6	0.1
			200	5.2	–1.3
	Urine		2.5	13.6	3.4
		1.5–500	250	7.9	0.4
			500	13.3	1.9
R/S-ketamine	Feces		2.5	7	–2.5
		1–1,000	500	3.3	–2.4
			1,000	5.4	1.4
S-norketamine	Serum		1.5	12.8	5.2
		0.5–200	100	11.5	–2.1
			200	6.5	–3.3
	Urine		2.5	6.2	5.4
		1.5–500	250	3.7	–0.5
			500	3.4	2.4
R-norketamine	Serum		1.5	3.1	6.7
		0.5–200	100	7.5	–0.6
			200	3.8	–5.2
	Urine		2.5	7.1	2.2
		1.5–500	250	6.6	–2.0
			500	9.0	–1.2
R/S-ketamine	Feces		2.5	7	–2.5
		1–1,000	500	3.3	–2.4
			1,000	5.4	1.4
S-dehydro-norketamine	Serum		1.5	13.4	3.5
		0.5–200	100	11.1	–3.2
			200	10.9	–2.6
	Urine		2.5	9.5	–4.6
		1.5–500	250	2.6	6.3
			500	11.1	7.7
R-dehydro-norketamine	Serum		1.5	7.0	–3.2
		0.5–200	100	4.8	–1.4
			200	13.1	0.8
	Urine		2.5	9.5	–4.1
		1.5–500	250	6.3	7.4
			500	9.8	7.0
2S,6S-hydrox-norketamine	Serum		3	10.8	–4.6
		1–200	100	14.3	–14.1
			200	13.3	–4.8
	Urine	1–200	3	14.2	–5.0
			100	11.7	–7.8
			200	18.5	7.0
2R,6R-hydrox-norketamine	Serum	1–200	3	8.9	19.2
			100	14.7	–10.9
			200	8.2	3.6
	Urine		3	12.6	–9.6
		1–200	100	12.0	–2.0
			200	8.6	7.1

Appendix 2. Exact *P* Values of Friedman Test with Dunn's *Post Hoc* Multiple Comparisons Test in Figure 2

Testing of Dose Dependence		Friedman Test (*P* Value)	Dunn's Multiple Comparisons Test (Adjusted *P* Value)					
			10 *vs.* 20 mg Oral Administration	10 *vs.* 40 mg Oral Administration	10 *vs.* 80 mg Oral Administration	20 *vs.* 40 mg Oral Administration	20 *vs.* 80 mg Oral Administration	40 *vs.* 80 mg Oral Administration
Norketamine/ketamine	R-stereoisomer	0.014	> 0.999	0.018	0.396	0.097	> 0.999	> 0.999
	S-stereoisomer	< 0.001	> 0.999	0.002	0.013	0.033	0.169	> 0.999
2,6-Hydroxynorketamine/ ketamine	R-stereoisomer	0.001	0.066	0.018	0.001	> 0.999	> 0.999	> 0.999
	S-stereoisomer	0.287	0.474	0.859	0.859	> 0.999	> 0.999	> 0.999
Dehydronorketamine/ ketamine	R-stereoisomer	0.005	> 0.999	0.004	0.097	0.203	> 0.999	> 0.999
	S-stereoisomer	0.010	> 0.999	0.644	0.342	0.644	0.342	> 0.999
2,6-Hydroxynorketamine/ norketamine	R-stereoisomer	0.070	0.337	> 0.999	0.825	0.170	> 0.999	0.463
	S-stereoisomer	0.003	0.396	0.719	> 0.999	0.004	0.018	> 0.999

Testing of iv–Oral Administration Differences		Friedman Test (*P* Value)	Dunn's Multiple Comparisons Test (Adjusted *P* Value)			
			5 mg iv *vs.* 10 mg Oral Administration	5 mg iv *vs.* 20 mg Oral Administration	5 mg iv *vs.* 40 mg Oral Administration	5 mg iv *vs.* 80 mg Oral Administration
Norketamine/ketamine	R-stereoisomer	< 0.001	< 0.001	< 0.001	0.011	0.003
	S-stereoisomer	< 0.0001	< 0.0001	< 0.0001	0.024	0.005
2,6-Hydroxynorketamine/ ketamine	R-stereoisomer	< 0.001	< 0.001	< 0.001	0.001	0.007
	S-stereoisomer	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Dehydronorketamine/ketamine	R-stereoisomer	< 0.001	< 0.001	< 0.001	0.011	0.001
	S-stereoisomer	< 0.001	< 0.001	< 0.001	0.002	0.005
2,6-Hydroxynorketamine/nor- ketamine	R-stereoisomer	0.028	0.113	> 0.999	0.016	0.901
	S-stereoisomer	0.005	0.663	> 0.999	0.022	0.061

iv, Intravenous administration.

Appendix 3A. Exact *P* Values of Friedman with Dunn's *Post Hoc* Multiple Comparisons Tests in Table 1

Testing of iv–Oral Administration Differences		Friedman Test (*P* Value)	Dunn's Multiple Comparisons Test (Adjusted *P* Value)			
			5 mg iv *vs.* 10 mg Oral Administration	5 mg iv *vs.* 20 mg Oral Administration	5 mg iv *vs.* 40 mg Oral Administration	5 mg iv *vs.* 80 mg Oral Administration
Renal clearance						
Ketamine	R-stereoisomer	< 0.001	< 0.001	0.016	0.376	0.011
	S-stereoisomer	0.001	0.001	0.002	0.256	0.017
Norketamine	R-stereoisomer	0.236	> 0.999	0.151	0.259	> 0.999
	S-stereoisomer	0.551	0.816	> 0.999	> 0.999	0.424
Dehydronorketamine	R-stereoisomer	0.023	0.663	0.003	0.333	0.424
	S-stereoisomer	0.032	> 0.999	> 0.999	0.061	> 0.999
2,6-Hydroxynorketamine	R-stereoisomer	0.001	0.001	0.002	0.223	0.928
	S-stereoisomer	0.002	> 0.999	> 0.999	0.259	0.016
Terminal half-life						
Ketamine	R-stereoisomer	< 0.001	> 0.999	0.259	< 0.001	< 0.001
	S-stereoisomer	< 0.001	> 0.999	> 0.999	0.005	0.067
Norketamine	R-stereoisomer	0.100	> 0.999	> 0.999	0.024	> 0.999
	S-stereoisomer	0.107	> 0.999	0.377	0.292	0.034
Dehydronorketamine	R-stereoisomer	< 0.001	0.002	< 0.001	< 0.001	< 0.001
	S-stereoisomer	0.049	0.151	0.084	0.016	0.199
2,6-Hydroxynorketamine	R-stereoisomer	0.003	0.011	0.011	0.005	0.003
	S-stereoisomer	0.034	0.113	0.151	> 0.999	> 0.999
Time of maximum plasma concentration						
Norketamine	R-stereoisomer	< 0.001	0.005	0.022	< 0.001	< 0.001
	S-stereoisomer	< 0.001	0.004	0.011	< 0.001	< 0.001
Dehydronorketamine	R-stereoisomer	< 0.001	0.013	< 0.001	< 0.001	< 0.001
	S-stereoisomer	< 0.001	0.013	< 0.001	< 0.001	< 0.001
2,6-Hydroxynorketamine	R-stereoisomer	< 0.001	0.084	0.476	0.044	< 0.001
	S-stereoisomer	< 0.001	0.001	0.004	< 0.001	< 0.001

iv, Intravenous administration.

Appendix 3B. Exact *P* Values of the Mann–Whitney U Test for Comparisons between the Respective S- and R-stereoisomers in Table 1

	Unit	5 mg iv	10 mg Oral Administration	20 mg Oral Administration	40 mg Oral Administration	80 mg Oral Administration
		S-stereoisomers *vs.* R-stereoisomers	S-stereoisomers *vs.* R-stereoisomers	S-stereoisomers *vs.* R-stereoisomers	S-stereoisomers *vs.* R-stereoisomers	S-stereoisomers *vs.* R-stereoisomers
Dehydronorketamine						
$AUC_{0-\infty}$	ng × h/ml	0.006	< 0.001	< 0.001	< 0.001	< 0.001
Maximum serum concentration	ng/ml	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Terminal half-life	h	0.025	0.015	0.046	< 0.001	< 0.001
Cumulative amount of ketamine and metabolites excreted into urine	µg	0.022	0.059	0.431	0.011	0.011
Renal clearance	ml/min	0.130	0.007	0.003	0.520	0.005
2,6-Hydroxynorketamine						
$AUC_{0-\infty}$	ng × h/ml	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Maximum serum concentration	ng/ml	0.330	0.213	0.146	0.191	0.022
Time of maximum plasma concentration	h	< 0.001	0.003	0.012	0.008	< 0.001
Terminal half-life	h	< 0.001	< 0.001	< 0.001	0.044	0.024
Cumulative amount excreted into urine with glucuronide	µg	< 0.001	< 0.001	0.003	0.059	0.044
Cumulative amount excreted unchanged into urine	µg	< 0.001	< 0.001	0.002	0.054	0.044
Renal clearance	ml/min	0.760	0.040	0.005	0.024	0.006

$AUC_{0-\infty}$, area under the concentration-time curve from zero to infinity; iv, intravenous administration.